Online Data Supplement for:

Human Cardiac Development in the First Trimester:

A High Resolution MRI and Episcopic Fluorescence Image Capture Atlas

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Table S1. Human Embryo Imaging by MRI and Episcopic Fluorescence Image Capture`

Estimated Gestational Age (weeks)	Carnegie Stage	Total Number Embryos Imaged	Imaging by MRI*	Imaging by EFIC*	EFIC and MRI
6 4/7	13	3	2 (2)	1 (1)	
6 6/7	14	4	3 (3)	2 (1)	1
7 1/7	15	3	1 (1)	2 (2)	
7 3/7	16	8	6 (6)	4 (3)	2
7 5/7	17	4	2 (2)	2 (2)	1
8	18	6	4 (3)	2 (1)	
8 2/7	19	5	3 (3)	2 (0)	
8 4/7	20	5	3 (3)	1 (0)	
8 6/7	21	4	2 (2)	2 (0)	
9 1/7	22	6	5 (3)	1 (0)	
9 3/7	23	4	3 (3)	3 (1)	2
Totals		**52 (42)**	**34 (31)**	**22 (11)**	

*Number of specimen yielding good imaging data indicated in parenthesis.

Table S2. MRI Acquisition Resolution

Carnegie Stage	Total Number Embryos	Voxel dimensions acquired by MRI (um)		
13	2	37x35x35	29x35x35	
14	3	29x37x37	33x36x36	37x37x37
15	1	33x35x35		
16	6	39x37x37	47x37x37	39x37x37
		53x52x52	53x52x52	61x43x43
17	2	41x35x35	39x39x39	
18	3	43x60x60	42x44x44	42x54x54
19	3	41x55x55	65x59x59	
		82x87x70		
20	3	45x54x54		
		67x67x62	67x67x63	
21	2	51x56x56	62x51x51	
22	5	57x57x57	60x59x59	64x63x63,
		68x78x78	63x63x63	
23	3	117x105x105	117x105x105	117x105x105

QUICKTIME MOVIE LEGENDS

A. Quicktime Movies of 2D Image Stacks

Movie 1: companion to Figure 2F, EGA 7 5/7 (CS 17))
This image stack shows embryo at an estimated gestational age (EGA) 7 5/7 week, Carnegie Stage (CS) 17 embryo. Imaging is in an off-axis sagittal plane. * = interventricular foramen, also known as the primary foramen, primary interventricular foramen, and bulboventricular foramen. Arrowhead designates septum primum, seen extending from cranial aspect of atrial roof and extending caudally to septate the atria. RA = right atrium; LA = left atrium; V = ventricular mass; RV = presumptive right ventricle; LV = presumptive left ventricle

Movie 2: companion to Figure 3F, EGA 8 weeks (CS 18)
This video stack represents EFIC imaging of an EGA 8 weeks (CS 18) gestation fetal heart. Imaging is in an off-axis coronal plane. Right and left sided ventricular chambers can be seen, connected by an interventricular foramen. Atrial septation is complete, and the septum primum is seen fused with the atrioventricular cushions. The inlet component of the ventricular septum is still open. * = septum primum; arrowhead indicates the still open inlet ventricular septum; RA = right atrium; RV = right ventricle; LV = left ventricle.

Movie 3: companion to Figure 3G, EGA 8 weeks (CS 18)
This video stack shows EFIC imaging of an EGA 8 weeks (CS 18) human embryo. Imaging is in a coronal plane. This clearly delineates that the ventricular outflow tract septum is closed while a portion of the inlet ventricular septum is open. * = complete atrial septum; first arrowhead indicates the left ventricular outflow tract; second arrowhead indicates the right ventricular outflow tract; RA = right atrium; LV = left ventricle; RV = right ventricle.

Movie 4: companion to Figure 4D, EGA 8 weeks (CS 18)
This 2D image stack from MRI imaging of an EGA 8 weeks (CS 18) human fetus. This stack is shown in an off-axis coronal plane. Arrowheads indicate thick right and left atrioventricular valves; asteriks denotes open inlet ventricular septum; arrow points to closed outflow tract septum. On either side of arrow, right and left ventricular outflow tracts can be noted. RA = right atrium; RV = right ventricle.

Movie 5: companion to Figure 4F, EGA 9 3/7 (CS 23)
2D image stack from MRI scan of an EGA 9 3/7 week (CS 23) human fetus. The imaging plane is in an off-axis coronal plane. Thinner appearance of the atrioventricular valves is seen. * = closed inlet septum; arrowheads point to the atrioventricular valves. Two crossing, distinct outflow tracts can be seen. RV = right ventricle; LV = left ventricle.

Movie 6: companion to Figure 5B, EGA 8 (CS 18)
2D image stack from MRI scan of an EGA 8 weeks (CS 18) human fetus. The imaging plane is in an off-axis coronal plane. Asteriks denotes open inlet ventricular septum; long arrow points to closed outlet septum. To the left of the arrow is the pulmonary artery, and to the right is the aorta. RA = right atrium; LA = left atrium.

Movie 7: companion Figure 5D-F, EGA 7 3/7 (CS 16)
2D image stack shows EFIC imaging of an EGA 7 3/7 week (CS 16) human embryo. The stack is shown in the transverse plane. The movie plays in the same sequence as truncal septation, i.e. from distal to proximal (see Figure 5 panels 5 F,E,D). Initially, the distal truncal outflow tract is

seen with lumens that exhibit a superior/inferior position. As the movie progresses, moving more proximally along the outflow, the aorticopulmonary septum is observed (denoted by an asterisk), with channels that exhibit right/left orientation. Most proximally, the outflow tract or truncus arteriosus is not yet divided. Below the truncus arteriosus, the presumptive right ventricular outflow tract can be seen. ** = interventricular foramen; RA = right atrium; LA = left atrium; RV = presumptive right ventricle; LV = presumptive left ventricle.

B. Quicktime Movies and QTVRs of 3D Reconstructions

Movie "a" is Quicktime movie of 3D model, and Movie "b" is interactive QTVR of the same 3D model.

Movies 8a,b: Companion Quicktime movie and QTVR to 3D model in Figure 3B (CS 14)
3D reconstruction of an embryo at EGA 6 6/7 weeks, CS14, shown in the frontal plane. Note the septum primum, with mesenchymal cap attached to the superior endocardial cushion. A common ventricular mass is seen.

Movies 9a,b: Companion Quicktime movie and QTVR to 3D model in Figure 3D (CS16)
3D reconstruction using EFIC imaging in the transverse plane of an embryo at EGA 7 3/7 weeks (CS 16). The developing ventricles are profiled. The presumptive left ventricular cavity can been seen, with interventricular foramen opening into the presumptive right ventricular cavity.

Movies 10a,b: Companion Quicktime movie and QTVR to 3D model in Figure 4C (CS16)
3D reconstruction using EFIC imaging of an EGA 7 3/7 weeks (CS 16) embryo. Imaging is in the sagittal plane. The developing atrioventricular valve structure is clearly delineated. The atrioventricular orifice is primarily positioned over the presumptive left ventricle, with cushions and supporting structures well seen. The presumptive right ventricle and truncus arteriosus, with two outflow cushions, are also profiled.

SUPPLEMENTAL FIGURES

Ten Figures Containing Enlarged Images of Thumbnails in Figures 7 and 8
The FIGURE NUMBER and PANEL LABELS correspond to panel labels of the individual thumbnails in Figures 7 and 8.

Figure S7A-E: Inflow

Figure S7F-J: Atrioventricular Junction

Figure S7K-O: Ventricular Mass

Figure S7P-T: Outflow.

Figure S8A-E: Atrial septation.

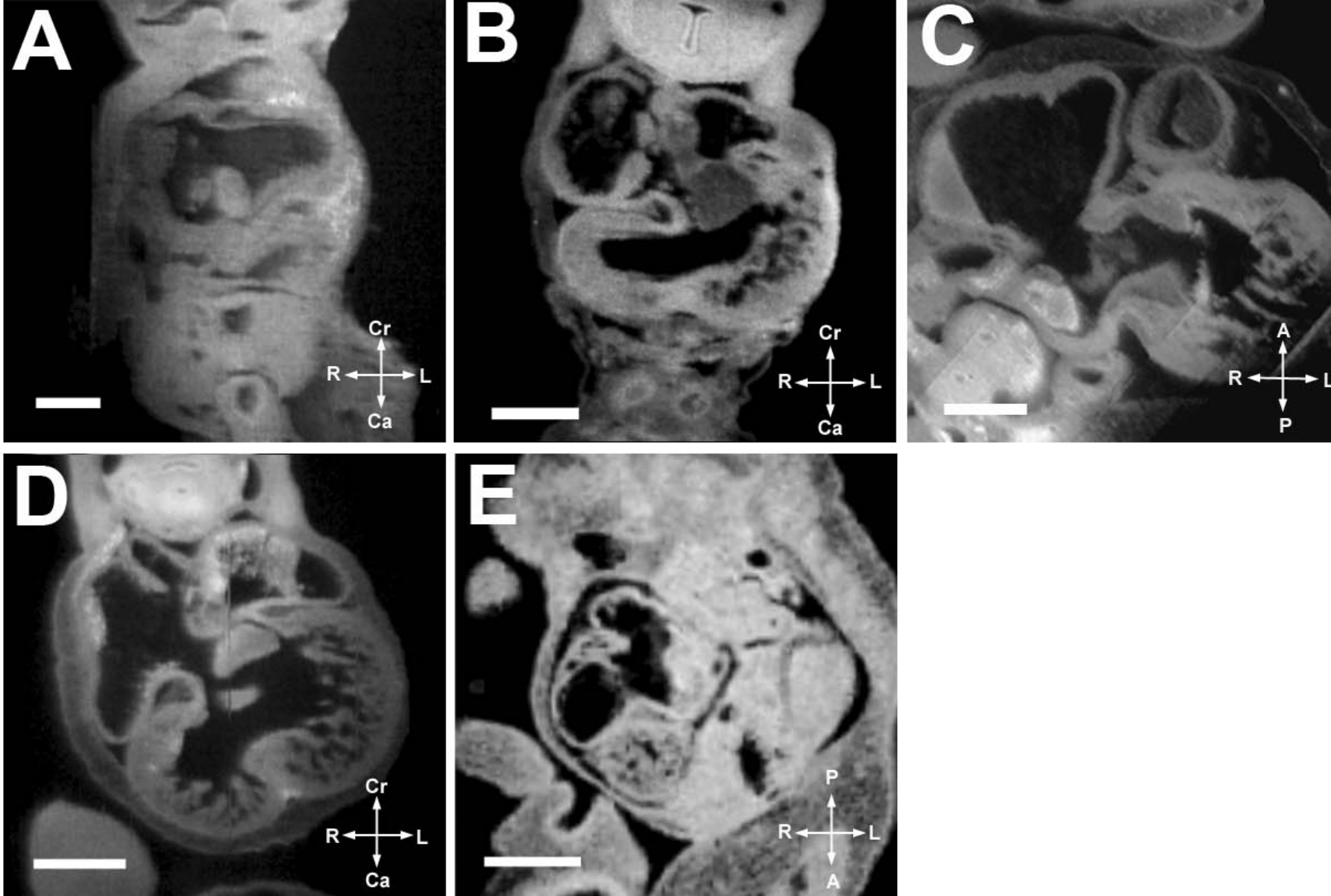
Figure S8F-H: Ventricular septation: Outflow Septum

Figure S8I,J: Ventricular septation: Inlet Septum

Figure S8K-O: Atrioventricular valves

Figure S8P-U: Semilunar valves: Aortic Valves

Figure S8V-AA: Semilunar valves: Pulmonary Valve



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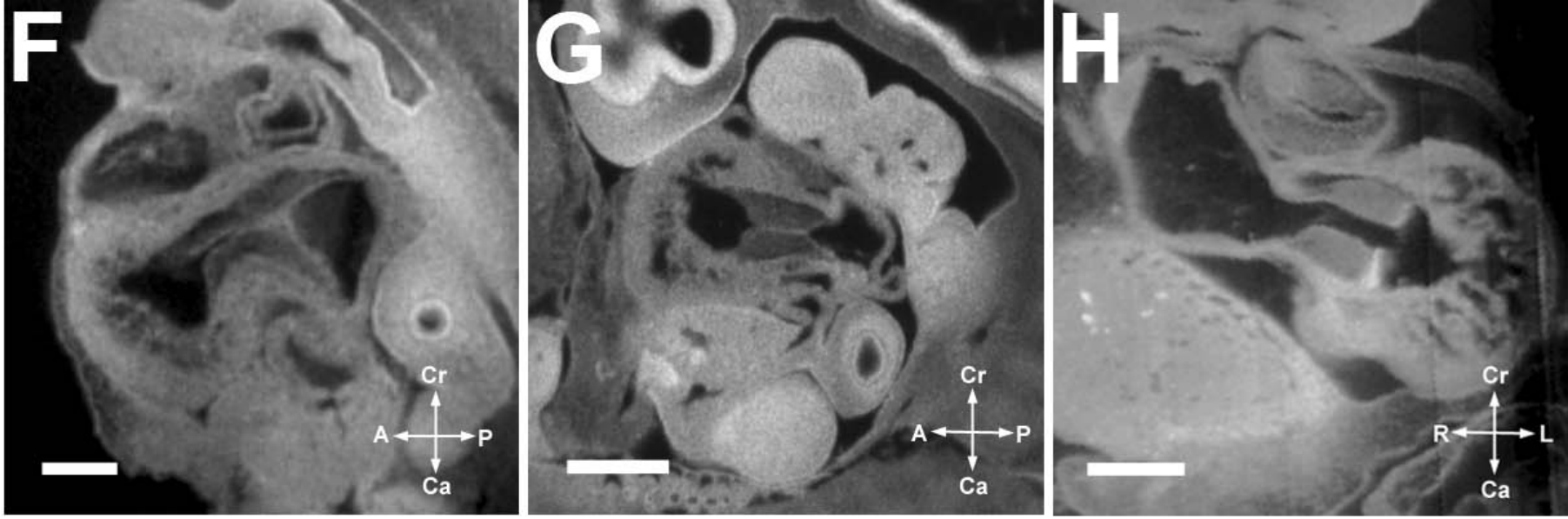

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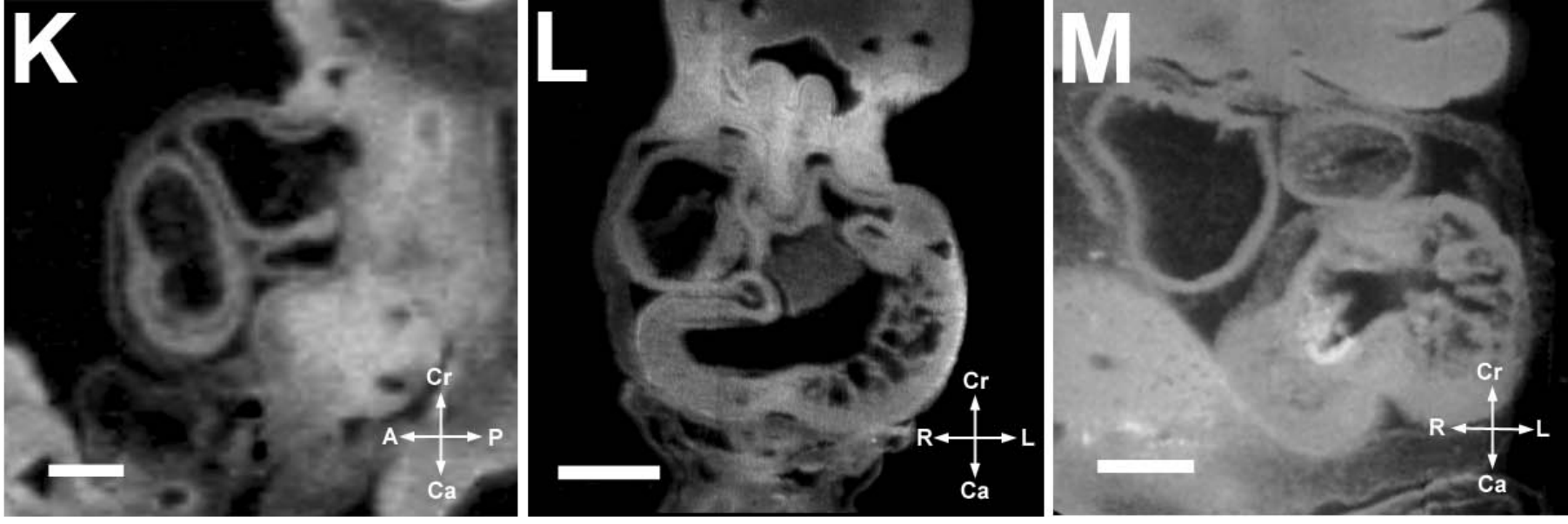

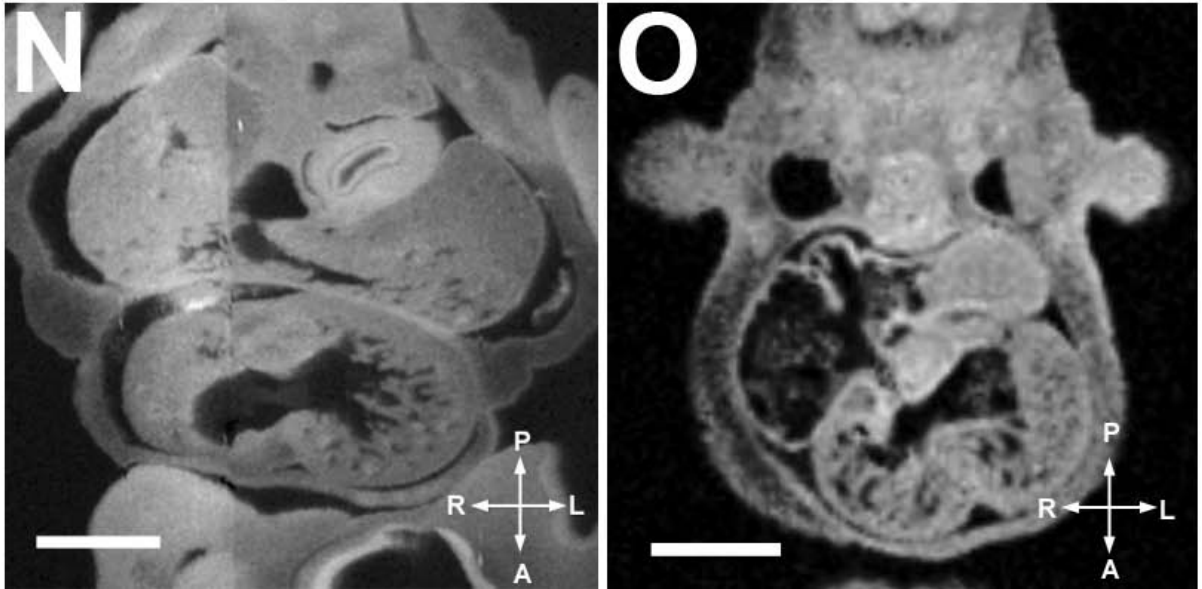

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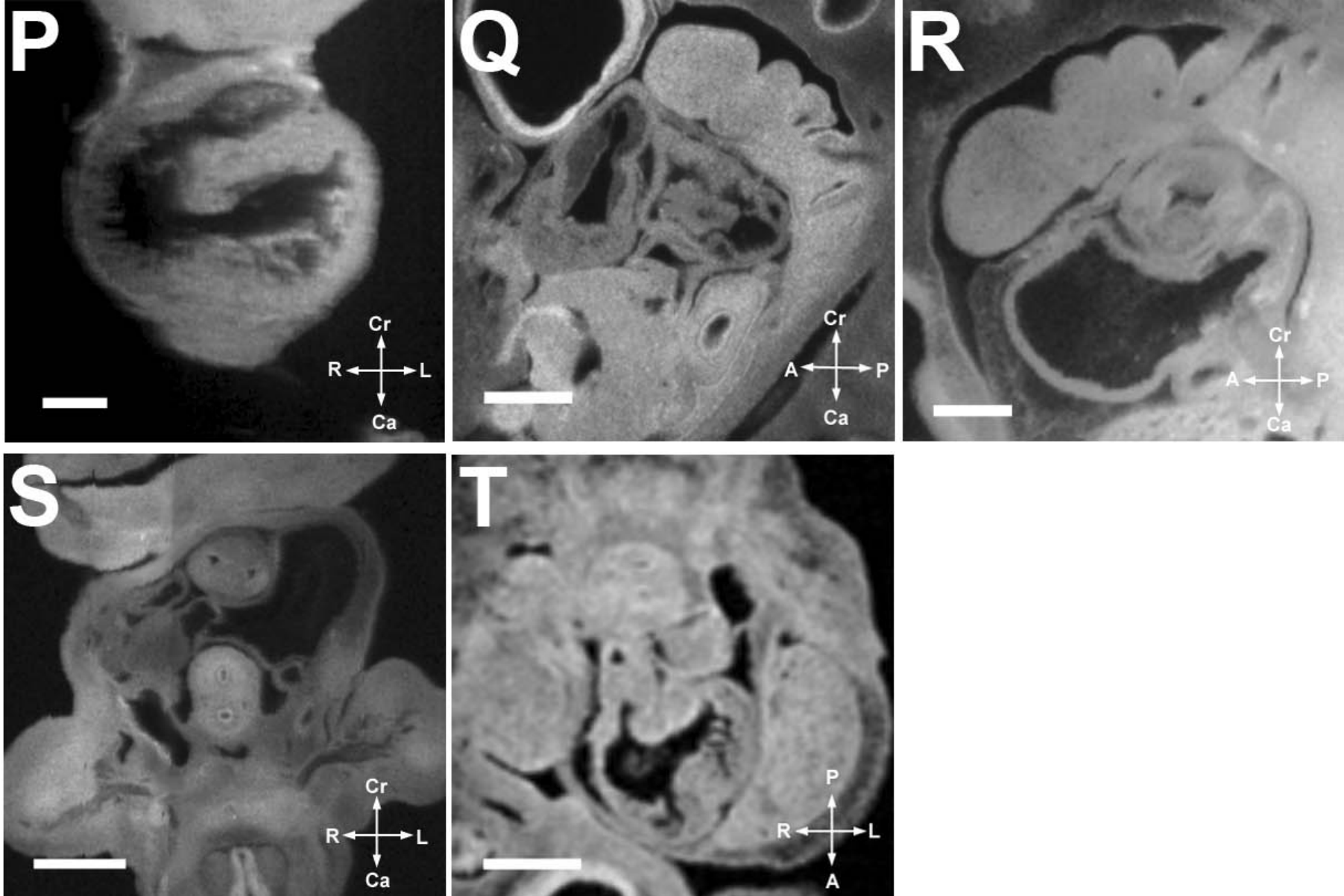

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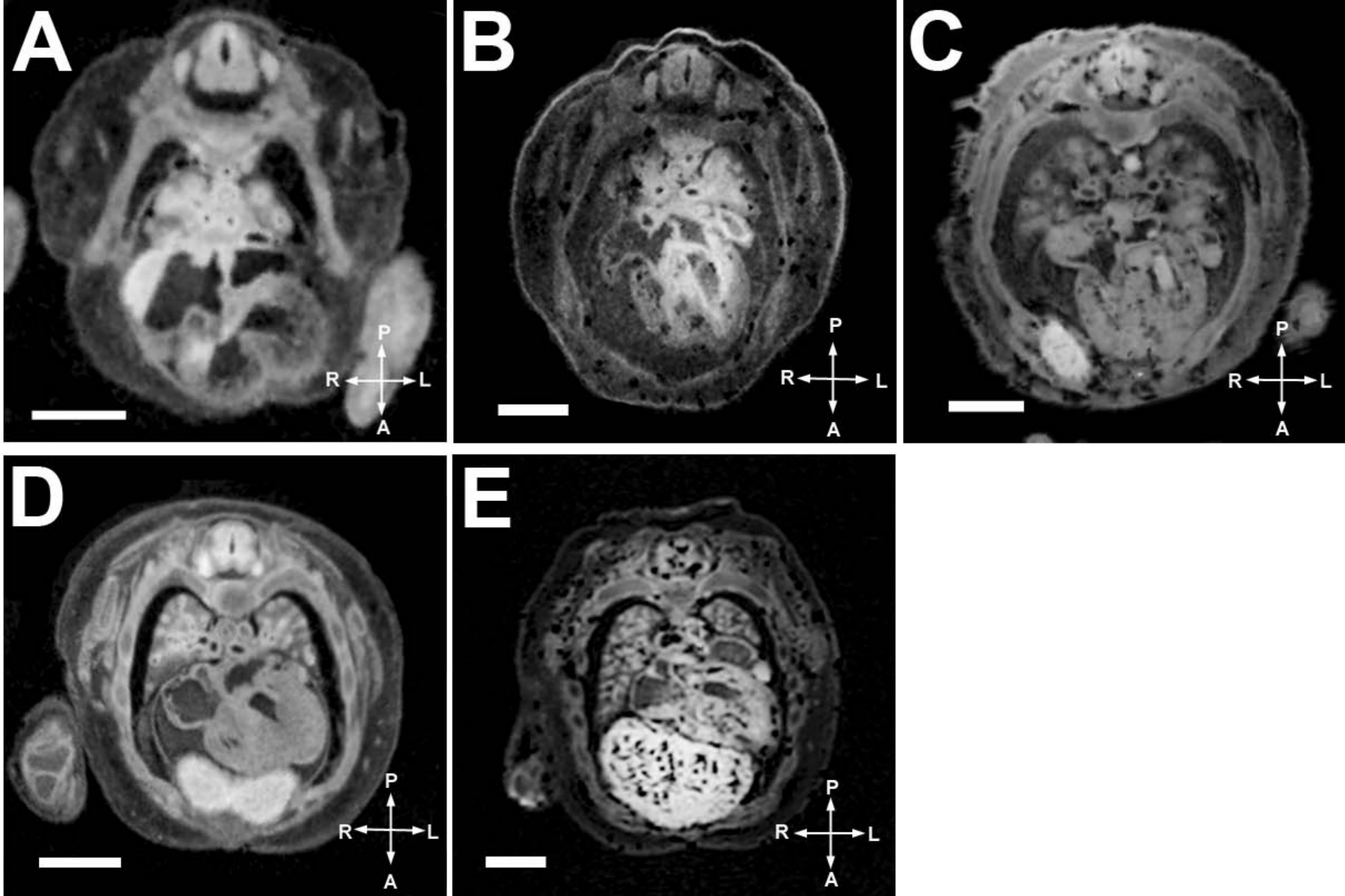

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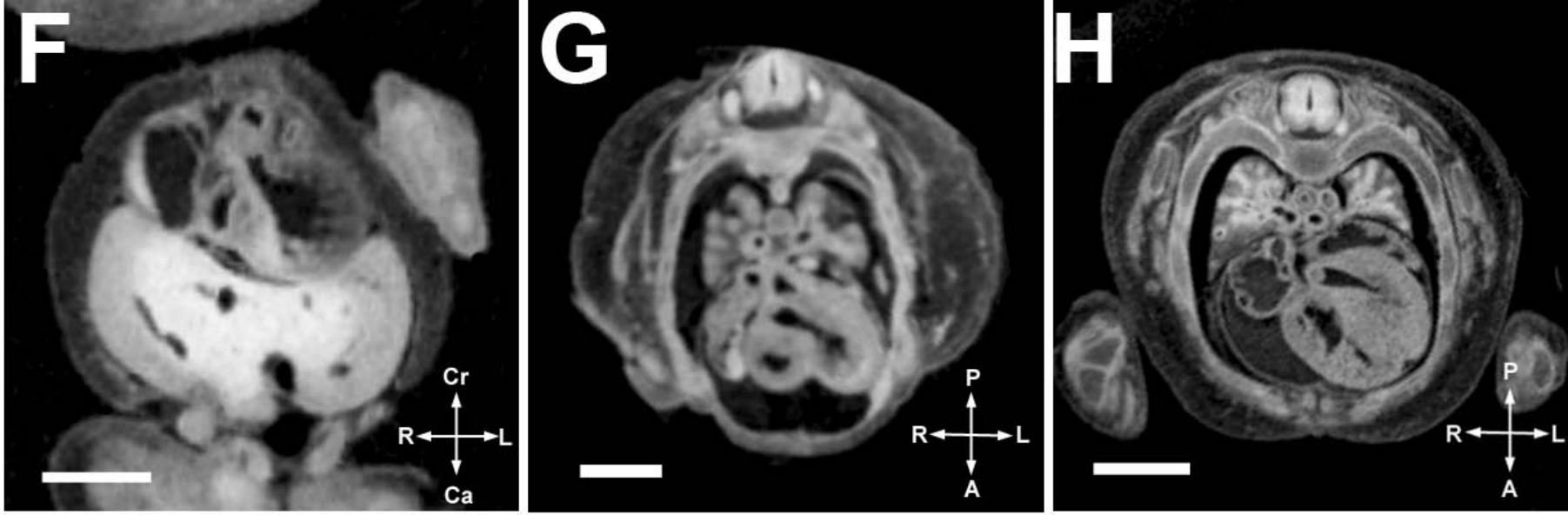

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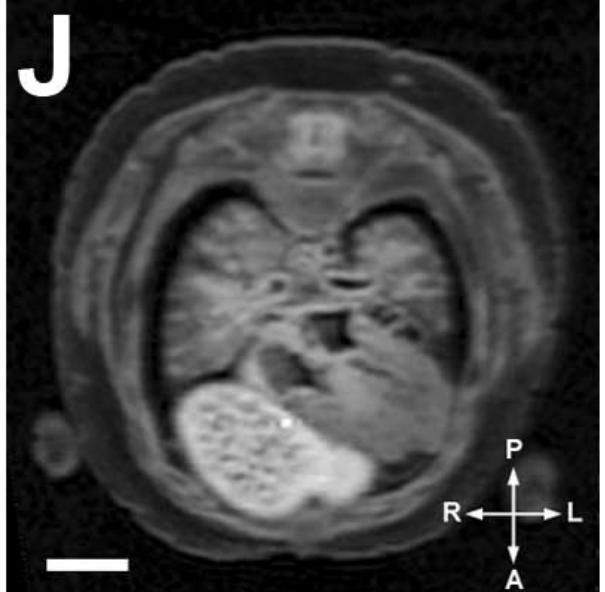

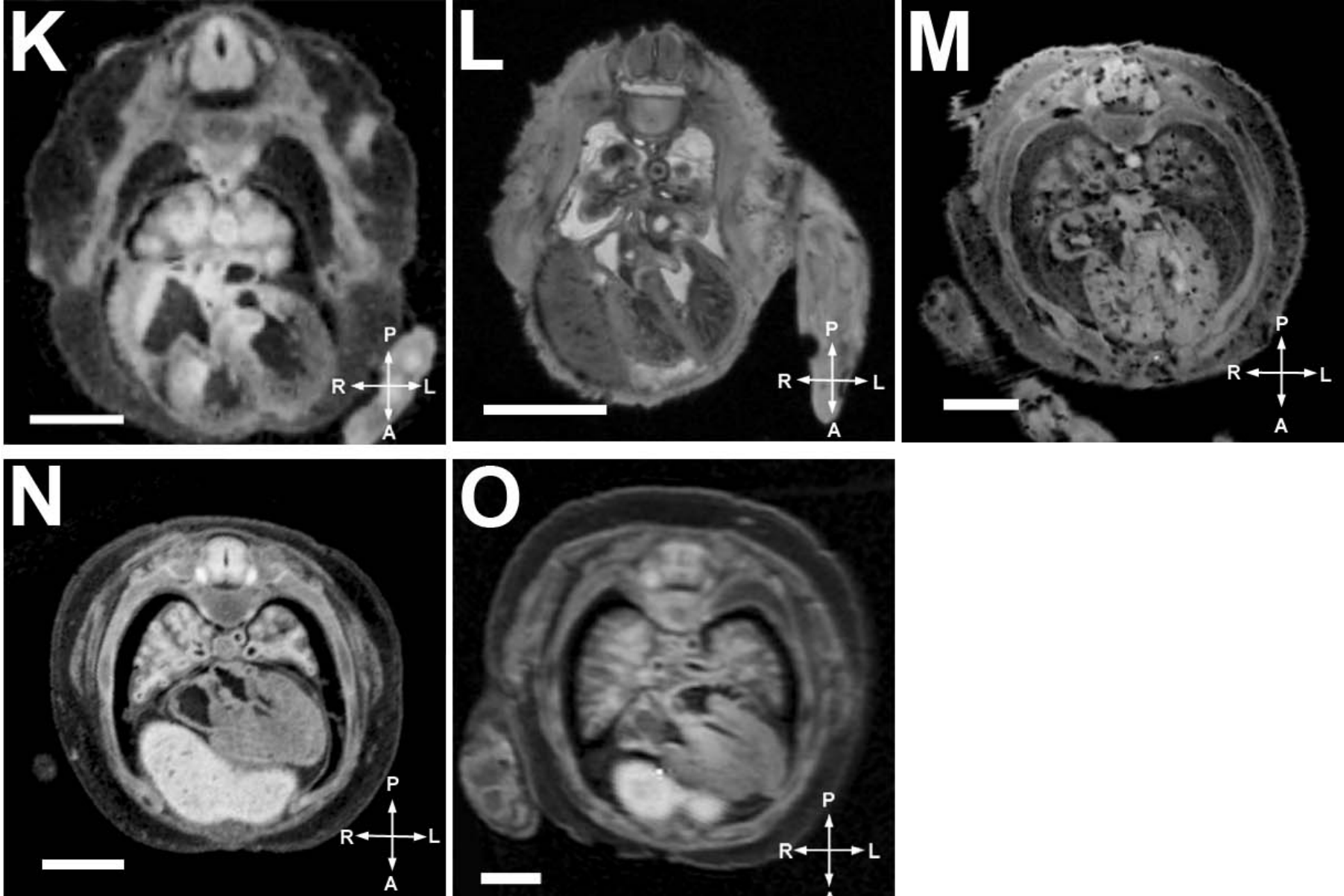

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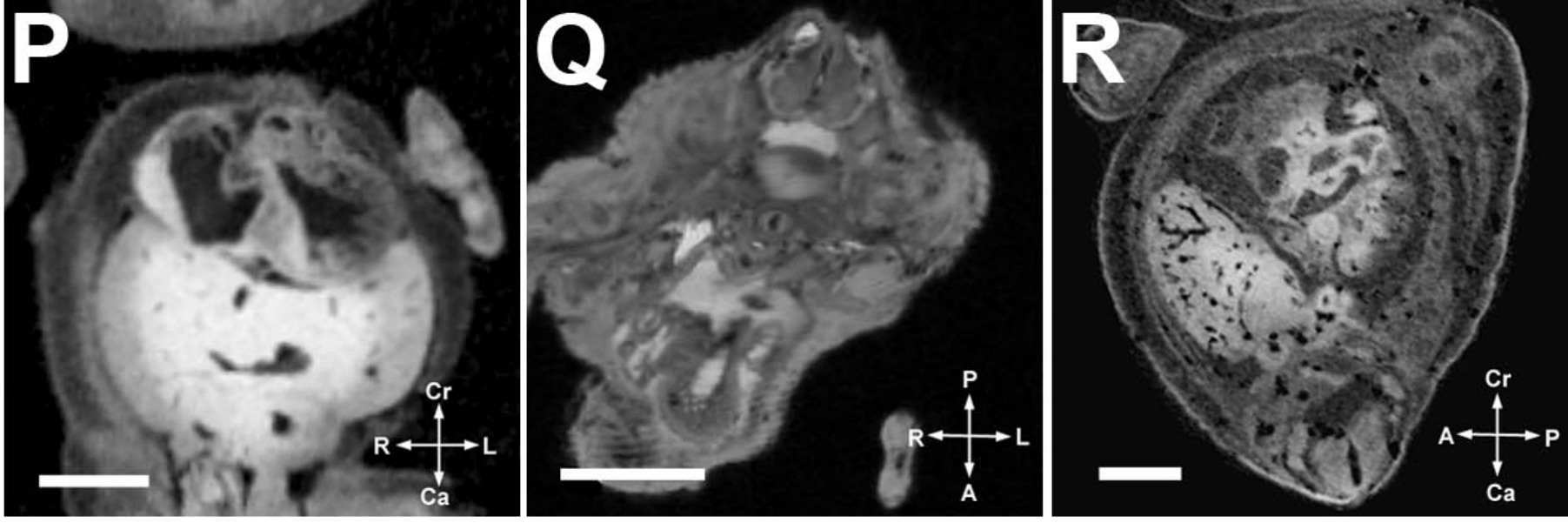

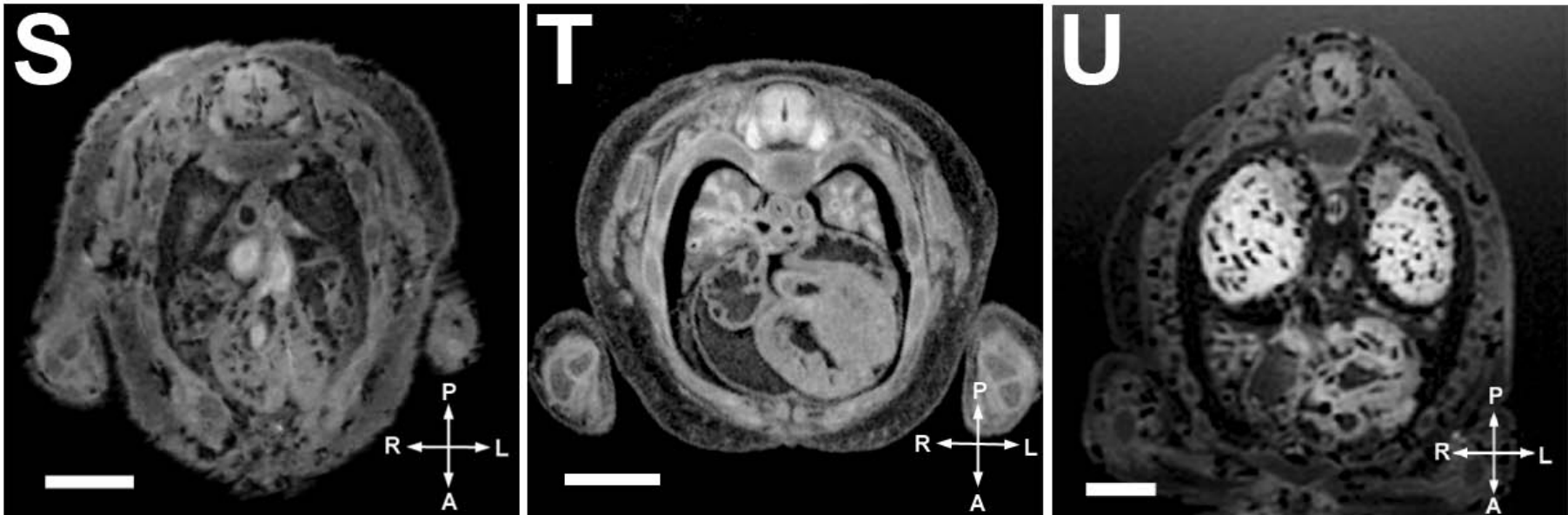

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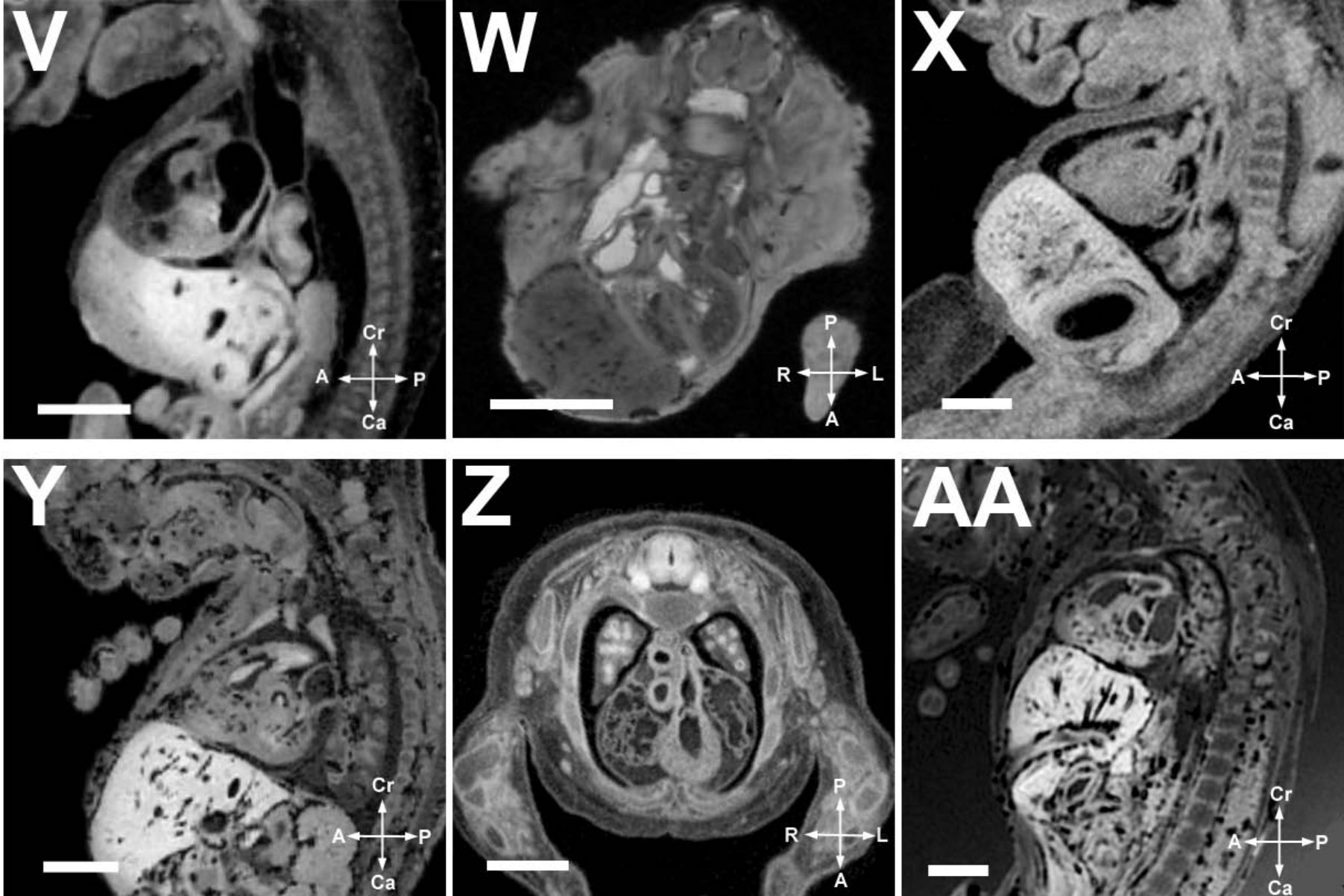

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